Production of Spike-in Controls for Gene Expression Assay Validation

Bioscience researchers are using DNA Microarrays as a powerful tool for inexpensive genome-wide experiments to survey the gene-based activity – the gene expression profile – of a biological system. The gene expression profile (which genes are active, at what magnitude) of a cell contributes strongly to its unique nature and properties, enabling different cells that contain identical genetic information to have a wide variety of properties. While it has great promise in genomic medicine (as a clinical diagnostic device, or for use in pharmaco- and toxico-genomics) the DNA microarray is currently limited to the "For Research Use Only" marketplace because there is no way to establish and demonstrate measurement quality. A NIST-hosted CRADA consortium, the External RNA Controls Consortium (ERCC), has been formed to address the need for validation of gene expression assays using microarrays and QRT-PCR experiments.

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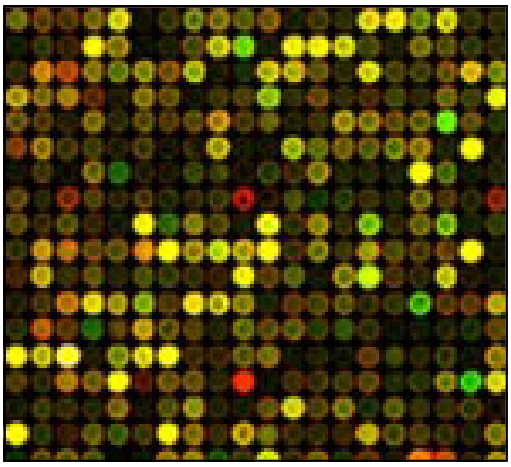
Approval of microarray gene expression for clinical use will require the ability to validate measurements and demonstrate that measurement results have sufficient sensitivity, specificity, reproducibility, robustness, reliability, accuracy, and repeatability. Estimating quantitatively these measurement performance figures-of-merit will require standards and methods that do not at present exist. Discordance of reported results from microarray studies, and difficulties in reproducing study results, has made it difficult to establish reliable correlation of a measured gene expression profile to a biological state or treatment outcome. The discordance and irreproducibility can only be addressed through a more complete understanding of the measurement, and is essential to support the move of this technology to the clinic.

A NIST-hosted CRADA consortium, the External RNA Controls Consortium (ERCC), has been formed to address the need for validation of gene expression assays using microarrays and QRT-PCR experiments. The ERCC is an ad-hoc group with approximately 50 members from academia, industry, and governmental organizations, initiated to develop a set of external RNA control transcripts that can be used to assess technical performance in gene expression assays. NIST, as host, plays a unique role in the organization providing "neutral ground" in which all the members can work equitably towards common goals. NIST also provides the necessary technical expertise to develop and provide well-characterized materials that will not only serve as controls, but also can be used to further understand the quality of what is being measured.

These materials include 100 well-characterized RNA transcripts that will be available as control materials, their use is supported through protocols for the preparation and use of the controls, and algorithms and bioinformatics tools suitable for their analysis. Other deliverables included in this program are access to clones of the reference set and publicly available sequence information and test data, thus enabling future use of the material and information in ways not currently envisioned.

The use of the ERCC materials will enable technical validation of gene expression experiments, allowing researchers to concentrate on investigation of biological variation. NIST is planning on the use of the ERCC controls for better understanding of gene expression measurement quality, specifically in the determination of the figures of merit, and is intended to accelerate application of this technology to diagnostic use. Regulatory calls for improved sufficient sensitivity, specificity, reproducibility, robustness, reliability, accuracy, and repeatability can only be answered through the application of well-characterized controls such as those produced by the ERCC.

http://www.cstl.nist.gov/biotech/workshops/ERCC2003/



A DNA microarray